

INSTRUCTIONS FOR INOCULATION OF BSK MEDIUM IN SUSPECTED CASES OF LYME DISEASE

MICHIGAN DEPARTMENT OF COMMUNITY HEALTH

NOTE: If the specimen container is received leaking, not properly labeled, test requisition not completed or the specimen label does not match the test requisition, the specimen will not be tested.

1. **Store un-inoculated tubes of media in the refrigerator @ 4° C until the expiration date shown on the tube.** Outdated media **must not be used** and must be discarded. Additional media can be obtained by faxing your order to: 517-335-9039 or by calling 517-335-9867.
2. Remove one tube per specimen to be inoculated from the refrigerator and warm to room temperature.
3. **Label tube** with the date inoculated, type of specimen and same name/unique identifier used on the test requisition. You will use it to link the specimen to the patient.
4. Place skin biopsy or 0.3 mL of sterile body fluid into warmed tube of media. **Hold specimen @ room temperature until ready to ship.**
5. When ready to ship, place no more than two tubes, wrapped in absorbent material provided, into aluminum screw-capped can and tighten the cap.
6. Place aluminum can **with completed test requisition for each specimen** on the outside of the container into screw-capped cardboard container and secure tightened cap with tape. **Note: A Lyme Disease Case Report Form (CDC 52.60) must be completed on each patient as well.**
7. Complete the white return mailing/Diagnostic specimen label and apply to cardboard container and ship by the most rapid and convenient means available (e.g., courier, bus, U. S. First Class, Priority or Express mail etc.).

NOTE: The shipper is responsible for being sure that their package is in compliance with the current shipping regulations.

See Reverse for Biopsy and Needle Aspiration Instructions

Biopsy and Needle Aspiration of Skin Lesions

Generally, the biopsy site is anesthetized (1 cc of a 1% lidocaine plus 1:100,000 epinephrine solution) and then disinfected (iodine tincture followed by isopropyl alcohol). Using a gentle twisting action, the punch instrument is used to cut the skin to a depth of 3 to 7 mm. The punch instrument is then removed. The skin punch is grasped with fine-tipped forceps, pulled gently away from the body and snipped at the base with iris scissors. Hemostasis is achieved in the usual fashion; generally, pressure alone is adequate. The biopsy specimen(s) can be divided with a scalpel if more than one test is planned. Biopsy samples are inoculated directly into room temperature culture medium.

A 2-needle, 2-person aspiration technique (Acutaneous lavage®) was developed in 1990-1991 by Gary Wormer, Gilda Forseter and colleagues at New York Medical College, based on a similar method described by Plesman et al (J Infect Dis 163:895:897, 1991) for use in rabbits. After local anesthesia and disinfection (as above), a 25 ga. 5/8" needle connected to a 3 cc syringe containing 2 cc of non-bacteriostatic normal saline is introduced intradermally and enough saline is injected to create a visible wheal at least 1 cm in diameter. While this first needle remains in place, a second operator introduces a 20 ga. 12" needle attached to a 3 cc syringe at a slanting angle into the wheal at a site opposite to that of the first needle. The second needle and syringe are then removed and used to collect fluid exuding from the second needle track. The rate and amount of fluid obtained from the second needle site depends on the rate and amount of additional saline injected into the wheal through the first needle and syringe. Aspiration fluid is inoculated directly into room temperature culture medium. The method seems to be less sensitive than culture of skin biopsy material.

A good location for isolation of spirochetes from EM lesions appears to be about 4 mm inside the perimeter (Berger et al., J Clin Microbiol 30:359-361, 1992).

CDC/DVBID
Lyme Disease Surveillance Summary
P.O. Box 2087
Fort Collins, CO 80521